

**CONGRUITY BACKCROSSING (CBC) AS A MEANS OF CREATING GENETIC DIVERSITY IN SEED COAT PHENOTYPES OF *PHASEOLUS VULGARIS* AND *P. ACUTIFOLIUS*.** Peter D. Ascher and Neil O. Anderson, Dept. Horticultural Science, Univ. of Minnesota, St. Paul, MN 55108

**Introduction.** Plant breeding programs have used different crossing methods to integrate economically important traits from wild species into cultivated crops. The most common system for interspecific gene transfer at the whole plant level is recurrent backcrossing (RBC) or introgressive backcrossing; others include inbred-backcross-line and recombinant inbreeding (Sullivan and Bliss, 1983; Thomas and Waines, 1984; Gepts and Bliss, 1985). With RBC, the direction of the initial cross uses the cultivated species as the maternal parent; the  $F_1$  primary hybrid and every subsequent generation is backcrossed to the cultivated parent. The goal of such a mating scheme is to incorporate the economically important trait from the wild species into the cultigen without any of the traits from the wild species that would be detrimental or unsuitable in cultivation. RBC has only infrequently been capable of such a task; frequent problems with RBC are loss of the important trait(s) from the nonrecurrent parent, difficulties in transferring traits that are quantitatively inherited, chromosome or genome elimination, and large linkage groups that are difficult to break (Stephens, 1961).

Parker and Michaels (1986) noted that a "bridge crossing scheme would facilitate the introgression of *P. acutifolius* germplasm into incompatible *P. vulgaris* lines". Haghighi and Ascher (1988) were the first to report the use of congruity backcrossing (CBC) as a useful means to create interspecific hybrid bridges with *P. vulgaris*, *P. acutifolius*. In this crossing scheme, the hybrids are backcrossed with each of the parent species in alternate generations. This eliminates the problems noted above with the many forms of RBC as both genomes are constantly incorporated and less--if any--traits are lost. Fertility, as measured by percent stainable pollen and seed set from non-manipulated flowers, increased with each CBC generation regardless if the recurrent parent was cultivated *P. vulgaris* or wild-type *P. acutifolius*, e.g. while the primary  $F_1$  hybrids (15.1-34.8%) had less percent stainable pollen than the parents (77.4-98.7%), it increased with each CBC generation ( $44 \pm 22.1$  -  $79.9 \pm 19.2\%$ ) (Haghighi, 1986; Haghighi and Ascher, 1988). Typically, higher percent pollen stainability and number of seeds per pod were found when the cytoplasmic (female) parent of the original cross was the last CBC parent. When CBC pedigrees contained equal dosages of the parents, the CBC hybrids were intermediate; unbalanced pedigrees favored the expression of the majority parent (Haghighi and Ascher, 1988). These fertile, CBC hybrids meant that important *P. acutifolius* traits could be recovered in any generation where the tepary parent had the higher dosage level. Flower colors were various shades of pink--uncharacteristic of any *Phaseolus* species--and seed coat coloration varied unpredictably. Inflorescences in early generations were intermediate, while advanced CBC hybrids had indeterminate inflorescences resembling those of *P. coccineus*; stems subtending such racemes twined around supporting stakes. Completely new characters also appeared after the second backcross generation (CBC2) and incongruity was reversed between the two species.

The objectives of this study were to analyze morphological characteristics of seed coats, from *Phaseolus* accessions created with CBC versus RBC. The assumptions employed for these interspecific hybridizations, when using essentially pure lines as parents, included:  $F_1$  hybrid intermediacy to both parents for all morphological phenotypes controlled by the nuclear genomes, maternal inheritance of seed phenotypes (color, shape, size, and patterning), homogeneity and lacking segregation until the  $F_2$  generation.

**Materials and Methods.** Parents for the species hybrids were wild *P. acutifolius* var. *acutifolius* PI 263590 or G40045 ( $A_{19}$ ), *P. acutifolius* var. *latifolius* PI 406622 ( $A_{10}$ ), and cultivated *P. vulgaris* 'Red Cloud' kidney ( $V_1$ ).  $A_{19}$  is from Sonora, Mexico; the native habitat for  $A_{10}$  is not known.  $V_1$  is a commercial kidney bean obtained from Lyng's Seeds (Modesto, California, USA); kidney beans of this type originated in the southern Andes of Peru and Argentina. Plants were grown under greenhouse conditions ( $45^\circ$  N, St. Paul, Minnesota, USA) with  $18^\circ/22^\circ$  C (day/night temperature) and long day photoperiods (0600-2200 HRS). CBC or RBC pedigrees were derived from crossing the parents reciprocally, i.e.  $V_1 \times A_{10}$ ,  $A_{10} \times V_1$ ,  $V_1 \times A_{19}$ , and  $A_{19} \times V_1$ . Parental dosage levels were denoted with superscript numbers following the parental identifier, such that two doses of  $V_1$  or  $BC_1$  in the cross  $V_1 A_{19}$  would be indicated as  $V_1^2 A_{19}$ , etc. All primary or RBC hybrids and early CBC

hybrids were embryo rescued (ER) when  $V_1$  was the maternal parent.

Seed morphological data were recorded for the parents and CBC and RBC generations. Since seed coat coloration and size are maternally inherited, the visible phenotypes are always lagging one generation behind in expression. For example, the seed coat of an  $F_1$  interspecific hybrid displays the actual phenotype of the previous generation, the maternal parent. In order to characterize the actual  $F_1$  phenotype, one would have to examine the  $F_2$  patterns (from selfing) or cross it as female with another genotype. Phenotypes recorded were seed coat (testa) color and pattern, shape, and size.

**Seed Phenotype Segregation Patterns.** A summation of the phenotypic segregation is as follows:

1. RBC produced seeds that resembled the maternal, recurrent parent. While seeds had additional genetic modifications from the nonrecurrent parent, most nonrecurrent traits were partially or completely lost by the RBC3.
2. RBC  $A_{10}V_1$  hybrids recovered the  $A_{10}$  phenotype, with no kidney shapes present. However, in one case a new *P. acutifolius* phenotype arose, i.e. striated, oval-shaped beans in the  $F_1$  of  $A_{10}^3V_1$ .
3. Early CBC generations resembled primarily the maternal phenotype.
4. CBC must be conducted at least to CBC5  $F_2$  before paternal traits surface. The  $V_1A_{19}$  CBC pedigree series produced primarily kidney types, although parental *P. acutifolius* types surfaced in the  $F_2$  or  $F_3$  generations of  $V_1A_{19}$  and  $V_1^3A_{19}^2$ , respectively.
5. Usually an imbalance in parental dosage favors the expression of transgressive segregants. The higher dosage parent predominates when an imbalance is present. Equal parental dosage generations are frequently intermediate or maternal.
6. Transgressive segregants arise for coloration, shape, and color patterns. Paternally-inherited kidney types, that were  $V_1$ -like in appearance, appeared from CBC5 ( $A_{19}^3V_1^4$ ) onwards.
7. New colors emerged with kidney beans in the  $F_2$   $V_1^2A_{10}$  (striated red/brown, black, white) and  $F_2$   $V_1^2A_{10}^2$  (red/brown, white). Plants in the  $F_2$  of  $V_1^3A_{10}^3$  segregated 2:1 for kidney:acutifolius phenotypes. RBC series for this pedigree produced all kidneys, with no new colors or *P. acutifolius* types.
8. As CBC continued, the kidney shape often was supplanted by Great Northern types (e.g.  $F_2$   $V_1^4A_{19}^5$ ) or square, *P. acutifolius* types (e.g.  $F_2$   $V_1^4A_{19}^5$ ,  $F_3$   $V_1^3A_{19}^2$ ). Flat, small kidney-shaped beans surfaced in the  $F_3$  of  $V_1^2A_{19}^2$ ; these matched a phenotype in the reciprocal series ( $F_1$   $A_{19}^3V_1^4$ ).
9. Segregation continues beyond the  $F_2$  generation for CBC hybrids, often as far as the  $F_{10}$ .
10. Paternal control of seed phenotypes was noted in some cases, for primary hybrids ( $V_1^1A_{19}^1$ ) or advanced CBC generations, although paternal transmission was not consistent.  $V_1A_{19}$   $F_2$  phenotypes resemble primarily the  $A_{19}$  paternal parent in shape and coloration. Thus, the  $V_1A_{19}$   $F_1$  had the  $A_{19}$  paternal phenotype rather than the  $V_1$  maternal.

#### Literature Cited.

- Gepts, P. and FA Bliss. 1985.  $F_1$  hybrid weakness in the common bean. *The Journal of Heredity* 76:447-450.
- Haghighi, K.R. 1986. Methods of hybridization of two bean species: *Phaseolus vulgaris* and *P. acutifolius*. PhD Dissertation, University of Minnesota. 107 pp.
- Haghighi, K.R. and P.D. Ascher. 1988. Fertile, intermediate hybrids between *Phaseolus vulgaris* and *P. acutifolius* from congruity backcrossing. *Sex Plant Reprod* 1:51-58.
- Parker, J.P. and T.E. Michaels. 1986. Simple genetic control of hybrid plant development in interspecific crosses between *Phaseolus vulgaris* L. and *P. acutifolius* A. Gray. *Plant Breeding* 97:315-323.
- Thomas, CV and JG Waines. 1984. Fertile backcross and allotetraploid plants from crosses between tepary beans and common beans. *J Heredity* 75:93-98.
- Sullivan, JG and FA Bliss. 1983. Recurrent mass selection for increased seed yield and seed protein percentage in the common bean (*Phaseolus vulgaris* L.) using a selection index. *JASHS* 108:42-46.